

SUMMARY

[0005] The disclosure is based upon an ultra-sensitive method for detecting multi-drug resistant bacterial colonization or infection in a subject. In addition, the disclosure relates to methods for the rapid detection and characterization of MDROs, bacteria, viruses, fungi, yeasts, and molecular, phenotypic, and host biomarkers associated with infectious diseases, through high density matrix gene testing in order to provide the most comprehensive, high resolution profile of colonization, infection, and resistance pattern in a test sample. In various examples the method includes isolating a nucleic acid sample from a biological sample obtained from the patient by amplifying the antibiotic resistant genes KPC, NDM, OXA, VIM, IMP, CTX-M and VanA by contacting the nucleic acid sample with one or more amplification primers that specifically hybridize with the each of the antibiotic resistant genes to provide an enriched nucleic acid sample; detecting the presence of the antibiotic gene by contacting the nucleic acid sample with one or more detection primers that specifically hybridize with the each of the antibiotic resistant genes to in the enriched nucleic acid sample; classifying the patient as having a multi-drug resistant bacterial colonization or infection when one or more of the antibiotic resistant genes are identified in the enriched nucleic acid sample; and providing a contact precautions recommendation for the patient having a multi-drug resistant bacterial colonization or infection. Optionally, the antibiotic resistant genes further include one or more of IMI, SME, GIM, SPM, NMC, SFC, SHV, TEM, BEL, VEB, GES, PER, SFO, BES, TLA, ACC, CMY, MIR, ACT, DHA, MOX, FOX, or CFE.

[0005a] Disclosed is a method of screening a patient for multi-drug resistant bacterial colonization or infection comprising: isolating a nucleic acid sample from a biological sample obtained from the patient; amplifying at least one antibiotic gene subtype from each of the KPC, NDM, OXA, VIM, IMP, CTX-M, and VanA antibiotic resistant gene families by contacting the nucleic acid sample with one or more amplification primers that specifically hybridizes to a substantially homologous region common to at least two antibiotic gene subtypes in each antibiotic resistant gene family to provide an enriched nucleic acid sample; detecting the presence of the at least one antibiotic gene subtype by contacting the enriched nucleic acid sample with one or more detection primer that specifically hybridizes to the substantially homologous region or to another substantially homologous region common to at

[00016b] Disclosed is a kit for determining whether a patient is colonized or infected with a multi-drug resistant bacterium comprising: a biological sample collection means; a plurality of sets of primers, wherein each set of primers specifically hybridizes to a substantially identical region common to at least two antibiotic resistant gene subtypes within one of each of the KPC, NDM, OXA, VIM, IMP, CTX-M, and VanA antibiotic resistant gene families; a control nucleic acid sample; and an instruction, wherein said instruction classifies said patient as being colonized or infected with a multi-drug resistant bacterium when at least one antibiotic resistant gene subtype from each of the antibiotic resistant gene families is identified in the biological sample.

[00017] Also disclosed is method for implementation by one or more data processors forming part of at least one computing system by receiving, by at least one data processor, data characterizing test results of biological specimens for a patient or group of patients, wherein the test results characterize an outcome of a nucleic acid sample contacted with one or more primers that specifically hybridize with one or more antibiotic resistant genes selected from KPC, NDM, OXA, VIM, IMP, CTX-M and VanA and optionally one or more of IMI, SME, GIM, SPM, NMC, SFC, SHV, TEM, BEL, VEB, GES, PER, SFO, BES, TLA, ACC,

[00026] Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure pertains. Although methods and materials similar or equivalent to those described herein can be used in the practice of the present disclosure, suitable methods and materials are described below. All publications, patent applications, patents, and other references mentioned herein are expressly incorporated by reference in their entirety. In cases of conflict, the present specification, including definitions, will control. In addition, the materials, methods, and examples described herein are illustrative only and are not intended to be limiting.

[00027] Other features and advantages of the disclosure will be apparent from and encompassed by the following detailed description and claims.

BRIEF DESCRIPTION OF THE DRAWINGS

[00028] Figure 1 is an illustration showing that the assay of the invention is capable of presumptively stratifying bacterial species based upon test results.

[00029] Figure 2 is a chart showing the limits of detection (CFU/swab) of the methods of the invention

[00030] Figure 3 is a chart showing the specificity of the methods of the invention.

[00031] Figure 4 is a chart showing the reproducibility of the methods of the invention.

[00032] Figure 5 is a summary of genes and gene families used in the Resistome Test.

[00033] Figure 6 illustrates the Resistome Test used as a first-line screen for establishing resistance profiles in combination with DNA sequencing for high resolution strain typing.

[00034] Figure 7 illustrates the utility of the Resistome Test used in combination with DNA sequencing data to establish gene transmission profiles and events.

[00035] Figure 8 illustrates an example of using the Resistome Test for reporting and mapping individual patient anatomical and point-source environmental data.

[00036] Figure 9 illustrates an example of using the Resistome Test for reporting and mapping multiple patient data across a hospital setting.

DETAILED DESCRIPTION OF THE INVENTION

[00037] The present invention relates to methods for the rapid detection of multi-drug resistant organisms (MDROs). The methods are useful in the detection of subjects that are

19. The method of claim 17, further comprising

recommending that the patient does not receive a carbapenem antibiotic when at least one antibiotic resistant gene subtype from the KPC, NDM, OXA, VIM, or IMP antibiotic resistant gene family is detected;

recommending that the patient does not receive a beta-lactam antibiotic when at least one antibiotic resistant gene subtype from the CTX-M antibiotic resistant gene family is detected; or

recommending that the patient does not receive vancomycin when at least one antibiotic resistant gene subtype from the VanA antibiotic resistant gene family is detected.

20. The method of claim 17, further comprising identifying one or more antibiotic resistant gene subtypes from at least one of the IMI, SME, GIM, SPM, NMC, SFC, SHV, TEM, BEL, VEB, GES, PER, SFO, BES, TLA, ACC, CMY, MIR, ACT, DHA, MOX, FOX, SIM, or CFE antibiotic resistant gene family, wherein

recommending that the patient does not receive a carbapenem antibiotic when at least one antibiotic resistant gene subtype from the SME, SFC, IMI, NMC, or CcrA antibiotic resistant gene family is detected;

recommending that the patient does not receive a cephalosporin antibiotic when at least one antibiotic resistant gene subtype from the PER, VEB, GES, BES, SFO, or TLA antibiotic resistant gene family is detected, a TEM antibiotic resistant gene subtype with amino acid substitution E104K, R164H, R164S, R164C, G238S, or E240K is detected, or an SHV antibiotic resistant gene subtype with amino acid substitution G156D, G238S, or E240K is detected; or

recommending that the patient does not receive a beta-lactamase inhibitor-beta-lactam combination when at least one antibiotic gene resistant subtype from the ACC, MOX, CMY, CFE, ACT, DHA, or FOX antibiotic resistant gene family is detected.

21. A surveillance method to identify a source of a multi-drug resistant bacterial infection or colonization, comprising:

obtaining a nucleic acid sample from a sample collected from the environment;

amplifying at least one antibiotic resistant gene subtype from each of the KPC, NDM, OXA, VIM, IMP, CTX-M, and VanA antibiotic resistant gene families by contacting the nucleic acid sample with a plurality of sets of amplification primers, wherein each set of amplification primers specifically hybridizes to a substantially identical region common to at least two antibiotic gene subtypes within one of the antibiotic resistant gene families to provide an enriched nucleic acid sample, wherein the nucleic acid sample is contacted by an internal amplification control nucleic acid prior to amplification; and

detecting the presence of the at least one antibiotic resistant gene subtype from each antibiotic resistant gene family by contacting the enriched nucleic acid sample with a plurality of sets of detection primers, wherein each set of detection primers specifically hybridizes to the substantially identical region or to another substantially identical region common to at least two antibiotic resistant gene subtypes within one of the antibiotic resistant gene families;

wherein the presence of the at least one antibiotic resistant gene subtype from each antibiotic resistant gene family in the enriched nucleic acid sample identifies the source of the infection or colonization.

22. The method of claim 21, wherein at least one antibiotic resistant gene subtype from one or more of the IMI, SME, GIM, SPM, NMC, SFC, SHV, TEM, BEL, VEB, GES, PER, SFO, BES, TLA, ACC, CMY, MIR, ACT, DHA, MOX, FOX, SIM, or CFE antibiotic resistant gene family is amplified and detected.